INTRASPECIFIC AGGRESSION AND ITS EFFECT ON THE DISTRIBUTION OF *ANTHOPLEURA ELEGANTISSIMA* AND SOME RELATED SEA ANEMONES

LISBETH FRANCIS

Biology Department, University of California, Santa Barbara, California 93106

Contiguous aggregations of the west coast sea anemone *Anthopleura elegantissima* are composed of individuals from a single clone, the products of asexual reproduction. In the field, adjacent clones of anemones are observed to be isolated from each other by anemone-free spaces; and in the laboratory a group of anemones of mixed clonal origins will reaggregate into isolated uniclonal groups. (Francis, 1973). In the field I have also observed that individuals of the large solitary form of *Anthopleura elegantissima* also remain isolated from adjacent members of the species. Contacts between genetically different individuals of this species initiates in one or both individuals an elaborate behavior pattern that usually results in damage to one or both animals. This is aggressive behavior according to the definition of Carthy and Ebling (1964), "An animal acts aggressively when it inflicts, attempts to inflict, or threatens to inflict damage on another animal. The act is accompanied by recognizable behavioral symptoms and definable physiological changes" (page 1). This behavior has never before been reported for this species, and my purpose here is to describe this specialized aggressive behavior and to investigate the relationship between the aggressive behavior of this and related species and the observed field distributions.

MATERIALS AND METHODS

Animals and collecting methods

In synthesizing a description of the aggressive response, I observed individuals of both the solitary and the aggregating form of the anemone *Anthopleura elegantissima* (most recently described by Hand, 1955). The animals were observed and/or collected intertidally on the California coast between Pacific Grove and Santa Barbara, California.

The specimens of *Anthopleura elegantissima* used in the experiments were small to large sized individuals (from 1 to 5 cm across the expanded oral disc) of the aggregating form and large individuals (from about 5 to about 8 cm across the oral disc) of the solitary form of the anemone. These animals were collected intertidally within ten miles of the Hopkins Marine Station at Pacific Grove, California and between Gaviota and Santa Barbara, California. The very small sub-adult anemones used in one set of experiments were collected at Arroyo Hondo, an area just south of Gaviota. These animals were collected in October of 1971 in a mussel bed where they were attached to mussels and interspersed pebbles. The specimens of *Anthopleura artemisia* and *Anthopleura xanthogrammica* were also collected intertidally near Hopkins Marine Station. Specimens of *Actinea equina* used in

experiments were collected near the Gatty Marine Laboratory, St. Andrews, Scotland and near the Plymouth Laboratory, Plymouth, England. These anemones were removed from the rocks on which they were found by gently working a thin spatula under the edges of the pedal discs.

Laboratory holding conditions

At the four marine laboratories at which this work was done (Hopkins, Santa Barbara, the Gatty, and Plymouth) the anemones were kept in bowls supplied with flowing sea water. The anemones were fed intermittently except during experiments, when food was withheld.

Conditions under which aggression was observed

Contact between two genetically different anemones, not mere proximity, appears to be the condition necessary for the initiation of the aggressive response.

In my early experiments anemones collected from different clones were brought into contact in the laboratory by first allowing some of the animals to attach to pieces of glass or stone, and then moving these portable animals to within contact distance of other anemones. More recently the anemones were stimulated to initiate the aggressive response by repeatedly touching the experimental animal on the tip of a tentacle with a tentacle excised from a non-clonemate. This was done in a manner designed to simulate natural contacts between anemones. Contacts between anemones and excised tentacles were as brief as possible (lasting less than a second), and sufficient time (2–3 minutes) was allowed for the retracted tentacles of the experimental animal to re-extend between successive contacts. A single set of repeated contacts was always restricted to the tentacles of one quarter section of the anemone. Stimulation was discontinued when the acrorhagi of the anemone began to inflate rapidly, signaling the beginning of the inflation stage of the aggressive behavior. If the behavior did not then proceed to completion, additional stimulations were given. The total number of stimulations needed to elicit a complete aggressive response (including the movement of application) is designated the aggressive threshold. Stimulation was discontinued if the reaction was not elicited within a preset number of contacts.

An excised tentacle was used for up to ten successive contacts with the same anemone, and the tentacle was kept in cool sea water between contacts. (I found for *Anthopleura elegantissima* that tentacles used for many more than ten intraspecific contacts and even those rubbed repeatedly and with considerable force among the tentacles and against the column of a non-clonemate were still completely effective at eliciting aggression upon contact with any non-clonemate. This was true as long as the surface of the tentacles was kept free of any superficial clinging material.)

PROCEDURES, OBSERVATIONS AND RESULTS

Anthopleura elegantissima

Description of the aggressive behavior. Although, as a matter of convenience, tentacles excised from another anemone were used to stimulate anemones during

most laboratory experiments, it was found that contact with any of the external surfaces of a non-clonemate including the column, uninflated acrorhagi, or the intact surface of the pedal disc elicited aggression in laboratory maintained anemones.

I have also observed the behavior occurring naturally in tidepools during daytime low tides, especially as the tide begins to rise, bringing cool aerated water into the stagnant pools and submerging anemones previously exposed to the air. Aggression is common among these expanded anemones as they are jostled together by the movement of the water.

The following description of the aggressive response of *Anthopleura elegantissima* is, then, a composite description based on my numerous observations of the behavior rather than a description of a single episode.

For convenience I have separated the aggressive behavior of *Anthopleura elegantissima* into five stages: (1) stimulation (or initiation), during which the tentacles of the two anemones repeatedly come into contact and withdraw from contact; (2) inflation, during which the acrorhagi commence to become turgid; and (3) the movement of application, during which the inflated acrorhagi are pushed toward the source of stimulation; (4) application of ectoderm, during which damage is inflicted on another anemone; and (5) recovery, during which the anemone returns to its normal posture. The various stages, which correspond roughly with those described by Bonnin (1964) for the aggressive behavior of *Actinea equina*, are illustrated in Figures 1 through 7 and are described in full below.

When an expanded anemone is moved toward and first makes contact with another expanded anemone, the tentacle tips are usually the first parts to make contact. Typically, if two clonemates meet in this way their tentacles will at first withdraw from the contact and will then re-extend. This may happen several times in succession until finally the tentacles cease withdrawing and remain more or less in contact. If the anemones then move toward each other, their tentacles interlace without apparent further interaction.

If anemones from different clones come within contact range of each other, the reaction is very different. Tentacle contact between anemones from different clones is followed by rapid withdrawal of those tentacles involved and often of some nearby tentacles as well. The retracted tentacles then slowly re-extend until contact is again made between the two anemones. The tentacles withdraw again from contact, and the process of contact and withdrawal is repeated until the next stage of the behavior begins. During the period of repeated tentacle contact the acrorhagi (also called marginal spherules) closest to those tentacles involved in the contacts often become visible as white (or rarely pinkish) spherules beneath the outer cycle of tentacles (Fig. 1c). In unaroused anemones the acrorhagi, which are more or less numerous depending on the individual, are often difficult to see as they are usually quite small and lie hidden beneath the tentacles and in the fosse (a channel between the collar of the column and the tentacles).

The inflation stage of the aggressive behavior begins as the tentacles of the aggressor withdraw from the area of stimulation after repeated contact with a non-clonemate. As the tentacles begin to re-extend after the last contact, the anemone often assumes a more upright posture (Fig. 3). This may be accomplished by contraction of the circular muscles of the column and/or relaxation of
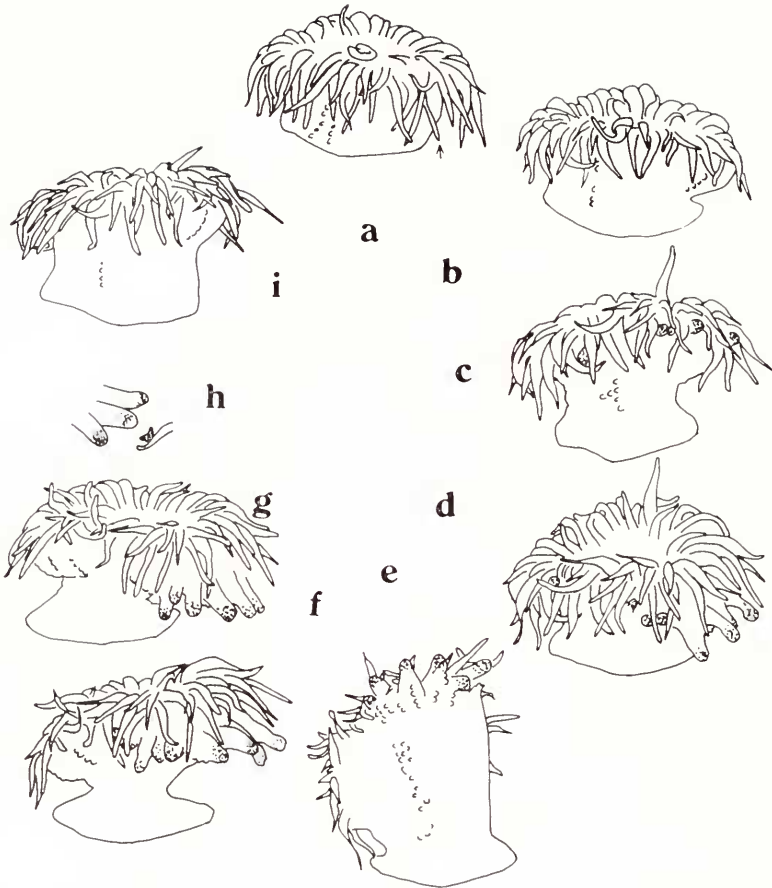
FIGURE 1. The positions of a specimen of *Anthopleura elegantissima* (the aggregating form) were traced from photographs taken during one aggressive episode. (a) The anemone is shown at rest (before contact with the non-clonemate); the arrow indicates the region that will come into contact with the non-clonemate. (b) Initiation: the anemone is shown just after contact with the non-clonemate; the relative retraction of the tentacles and a slight swelling of the capitulum (to the right) may be noted. (c) Inflation: the white tips (stippled) of the inflating acrorhagi have become visible in the area adjacent to the tentacles receiving the stimulation; the shape of the column is notably different from that shown in Figure 1a. (d) Inflation: the acrorhagi in the area of stimulation are strongly inflated, appearing white at the tips (stippled) and transparent at the bases. (e) Movement of application: the inflated acrorhagi are drawn upward and back away from the source of the stimulus. (f) Movement of application: the acrorhagi are sweeping downward and may, as in this case, continue downward to the position shown in Figure 1g, or the movement may stop in the position shown here with the anemone leaning over the adjacent non-clonemate. (g) Movement of application: the acrorhagi have reached the bottom of their downward sweep and are being wiped against the body of the victim. (h) Release of ectoderm: acrorhagial ectoderm has been released from one of the acrorhagi which is now transparent (unstippled) at the tip where the ectoderm is missing; the ectoderm is shown clinging to a tentacle of the victim. (i) Recovery: the acrorhagi have deflated but the anemone has not yet fully recovered its preaggression posture; the column is still somewhat elongated and the capitulum is still swollen and somewhat elevated in the area proximal to the area of stimulation.

the longitudinal muscles. There is usually also some swelling of the capitulum at the base of the stimulated tentacles, perhaps the result of relaxation of the circular muscles just below the stimulated tentacles and contraction of those muscles below the remaining tentacles. At this time the acrorhagi associated with the stimulated tentacles become distended, changing in appearance from shriveled white spherules to transparent rounded cones with white tips (Fig. 1d). The gastrovascular cavity of these anemones extends into the hollow tentacles and acrorhagi, and it is possible that the contractions of the circular muscles described above serve to force water from the columnar portion of the gastrovascular cavity into these acrorhagi, causing them to become distended.

Sometimes there are, in addition to the above movements, peristaltic contractions that begin at the base of the elongated anemone and travel up the column. These muscular contractions (probably of the circular muscles) may serve to force water into the expanding acrorhagi.

Along with an increase in the turgidity of the involved acrorhagi during the inflation period, there is usually a progressive increase in the number of acrorhagi that are inflated. Recruitment progresses in both directions around the capitulum so that more and more acrorhagi on either side of the originally stimulated tentacles become involved. Typically four to twelve acrorhagi become fully turgid during a single aggressive episode.

Acrorhagial inflation has never before been reported for this species; and except in rare, unexplained cases full inflation of the acrorhagi seems to occur only during the aggressive response.

The movement of application is a rather rapid movement for an anemone, requiring between 30 and 120 seconds for the complete movement.

As the expanding acrorhagi begin to distend, the swollen edge of the capitulum is extended upward drawing the turgid acrorhagi upward and away from the original site of contact with the other anemone (Fig. 4). This movement probably involves elongation of the longitudinal columnar muscles adjacent to the area of acrorhagial inflation. The anemone may remain in this position for a few seconds. Then the area of the oral disc between the mouth and the stimulated tentacles begins to elongate. This might be effected by relaxation of radially arranged muscles of the oral disc in this region and contraction of other muscles of the column and oral disc which would cause additional gastrovascular fluid to be forced into the area. As the oral disc changes shape the swollen edge of the capitulum moves downward, and the expanded acrorhagi point outward or somewhat downward (Fig. 5). This motion, which is perceived as a smooth downward sweep of the expanded acrorhagi, sometimes stops as the acrorhagi reach the position shown in Figure 1f (the aggressor is leaning over the spot where the victim normally should be). More often, the downsweeping movement continues until the acrorhagi are on a level with the pedal disc (Fig. 6) or actually touching the substratum. During this time inflation of the acrorhagi often continues, and more acrorhagi at both edges of the inflated area continue to be recruited.

The full movement of application sequence may be repeated one or more times, or the anemone may slowly recover its normal posture after only one movement of application.

If during the downward motion of the movement of application the white tip of one or more of the aggressor's fully inflated acrorhagi (*i.e.*, acrorhagi so turgid
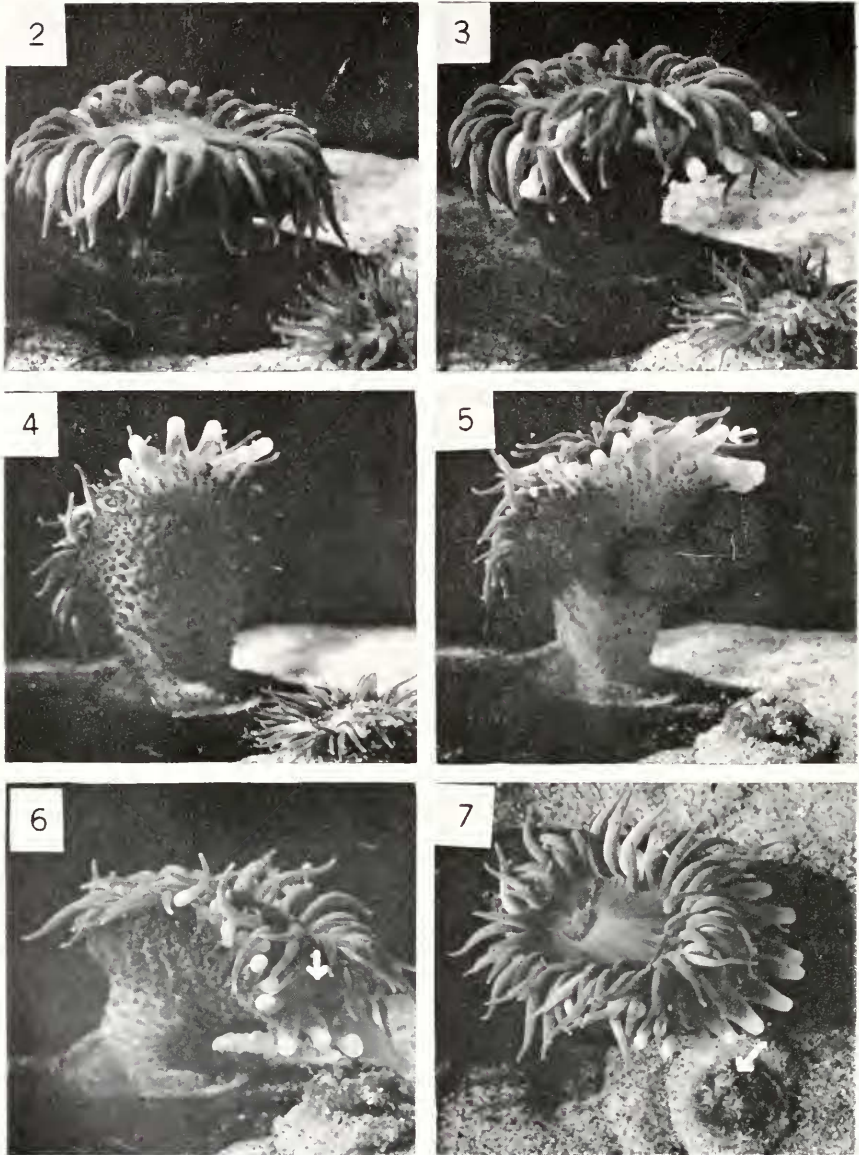
FIGURE 2.   The anemone is shown at rest.

FIGURE 3.   Inflation: the white tips of the inflating acrorhagi have become visible; the column has become more elongated.

FIGURE 4.   Movement of application: the fully inflated acrorhagi are drawn upward and back and are just about to begin moving downward toward the victim.

FIGURE 5.   Movement of application: a second movement of application is underway and more fully inflated acrorhagi have been recruited (cf. Fig. 4); transparent areas are obvious at the tips of several acrorhagi (→) where acrorhagial ectoderm was released from the acrorhagi and applied to the victim during the first movement of application.

that their bases appear transparent, see Fig. 7) comes into contact with a non-clonemate, the white surface layer can be observed to pull loose from the acrorhagus and adhere to the body of the non-clonemate (Fig. 1h and 7).

Fully expanded acrorhagi that had released part of their white surfaces as described above were sectioned using freeze sectioning and standard paraffin embedding techniques. In the region from which the white material was released the entire ectoderm layer was absent, and only the endoderm and mesogleal layers remained. *In vivo*, the tips of such acrorhagi appear transparent where the ectoderm is missing (Fig. 5).

During the slow return to a normal posture following the aggressive response, the anemone first rises from the arched over position (Fig. 1g) to an upright posture. The acrorhagi frequently lose their turgidity as the anemone returns to the upright position (Fig. 1i). If the anemone does not again come into tentacle contact with a non-clonemate, the acrorhagi eventually deflate and within a matter of hours the animal slowly returns to its more usual posture.

The anemone to which acrorhagial ectoderm has been applied (the victim of an aggressive episode) may respond by returning the aggression; more frequently, however, it will at first retract its tentacles, shorten its column, and contract the large sphincter at the top of the column. The damaged anemone may then move away from the aggressor; or sometime, after re-extending the tentacles and column and relaxing the sphincter, it may lean away, thus avoiding contact that would trigger another attack by the aggressor.

In other cases, apparently after having had insufficient time to attach its foot to the substratum or after having received severe damage as a result of repeated attacks, the victim may release its hold on the substratum. In the field, such an animal would certainly be carried away by the waves and might conceivably resettle elsewhere. In the laboratory, animals that have released their foothold after being attacked and damaged by a non-clonemate have subsequently resettled and recovered when they were removed to a separate bowl.

The tissue of the victim in the area of applied acrorhagial ectoderm shows obvious signs of damage and deterioration within about a day. This is especially evident for the tentacles, which contract and remain contracted as soon as acrorhagial ectoderm is applied. Within a few days, the obviously necrotic tissue in the damaged area is sloughed off along with the whitish patches of foreign acrorhagial ectoderm.

Tentacles to which acrorhagial ectoderm was applied were fixed, embedded in paraffin, sectioned, cleared, and stained (Masson's Trichrome dyes). In these sections it was possible to see the threads of the large atrich nematocysts (in this species, peculiar to the acrorhagi) penetrating the tentacle tissue.

As the following experiment demonstrates, an anemone that is repeatedly attacked and that can neither move away nor be washed away from the vicinity of an aggressor will eventually be killed.

FIGURE 6. Movement of application: the acrorhagi have reached the bottom of their downward sweep and are being wiped against the body of the victim; swelling of the fosse area between the inflated acrorhagi and the tentacles is apparent here ($\rightarrow$).

FIGURE 7. Movement of application: seen from overhead as the acrorhagi reach the level of the victim, elongation of the oral disc in the area proximal to the inflated acrorhagi is apparent; scraps of applied acrorhagial ectoderm are visible on the body of the victim ($\rightarrow$).

A single anemone from one clone (Clone 2) was first allowed to settle on a small pebble and then placed in a bowl so that it was close to and completely surrounded by anemones from another clone (Clone 1). The Clone 2 anemone was repeatedly attacked and damaged. Its pedal disc quickly became detached from the substratum (see Response of the victim, above); and being unable to escape the continued attack, it was killed within ten days. This experiment was repeated seven times, and each time the single Clone 2 animal was eventually killed.

As a control, one of two Clone 2 anemones placed in a bowl of Clone 1 anemones was protected from contact by a four-layered cylinder of plastic screening; the pebble on which the anemone was settled was the bottom of the cylinder. The tentacles of the protected Clone 2 anemone touched the inside layer of the screen, and the tentacles of the adjacent Clone 1 anemones touched the outside layer a fraction of a centimeter away. As a result, there was no aggression directed toward the protected Clone 2 anemone, and it remained undamaged and healthy in appearance for the several weeks that it remained in the dish. Within the same period the unprotected Clone 2 anemone was attacked, damaged, and killed.

Specificity of the aggressive response. In order to determine the range of circumstances in which *Anthopleura elegantissima* employs the aggressive response, the anemones were brought into contact with various animals that they might or might not encounter in their natural habitat. Both casual contact and repeated stimulation (described above under methods) were always used in attempting to elicit a response from the anemones.

Some of the invertebrates used were those that I have observed in the laboratory or in the field either feeding on *A. elegantissima* ("predator species") or being eaten by these anemones ("prey species"). Included among the predator species were the nudibranchs *Hermissenda crassicornis* and *Aeolidia papillosa*, the snails *Calliostoma annulatum* and *Epitoneum* sp., and the omnivorous batstar *Patiria miniata*. The two prey species used were young mussels *Mytilus* sp., and the surface-living coelenterate *Velella velella*. The anemones did not aggress after repeated contact with any of the predator or prey species, nor would the fully expanded acrorhagi of anemones in full aggression release ectoderm upon contact with any of these species.

The tentacles of numerous coelenterates and a chain of eudoxids from a calycophoran were also used in attempts to elicit aggression in *A. elegantissima*. These results were not always consistent. I observed a tendency for the tentacles of the two animals to stick together. Afterwards, the tentacles of *A. elegantissima* involved in the contact often looked shriveled and failed to react in the usual way to contact with the tentacles of a non-clonemate. Tentacles recently involved in intra-specific contacts only never appeared to be damaged. If, as seems apparent, damage is involved in at least some inter-specific contacts, this could cause some interference with the initiation of aggression. For this reason, each of the species of coelenterates tested was used for two to four series of stimulations and was recorded as eliciting aggression if even one series of contacts elicited the full aggressive behavior. None of the three Hydrazoans tested (the unidentified calycophoran, *Velella velella* and *Corymorpha palma*) was capable either of eliciting aggression or of triggering the release of acrorhagial ectoderm upon contact with the fully expanded acrorhagi of an aggressing anemone. All eight species of Anthozoans tested (*Anthopleura xanthogrammica, Anthopleura artemesia, Tealia* sp., *Cnidopus ritteri,*

Epiactis prolifera, Cerianthis sp., *Corynactis californica, Balanophyllia elegans* and *Paracyanthis stearnsii*) elicited aggression in *Anthopleura elegantissima* as well as triggering the release of acrorhagial ectoderm.

Inert objects were also used in an attempt to elicit the aggressive response. Aggression was not elicited by repeated tentacle contact with inert objects such as a clean glass rod or metal probe. Mechanical stimulation such as pinching the tentacles with a pair of forceps was also ineffective as was repeated electrical stimulation of the tentacles.

The following experiment was designed to determine whether the anemones aggress against all non-clonemate members of their own species or whether there might be clones that are mutually compatible.

Anemones from twenty-five different clones of the aggregating form of *A. elegantissima* and a large individual of the solitary form were collected from several different areas for use in this experiment. Several anemones from each of three clones (Clones 1, 2, and 5) were used as test animals in this experiment. Because it might be possible for an anemone to wander some distance from its clonemates and form a separate but genetically identical clone, care was taken when collecting in the vicinity of Clones 1, 2, and 5 to select only clones having color patterns observably different from those of the three test clones.

In the laboratory, securely settled anemones from each of the three clones were stimulated in the usual way with tentacles from each of the twenty-six genetically different anemones mentioned above. This included one set of forty stimulations using, as a control, tentacles from a clonemate not recently in contact with the experimental animal.

Because the members of the three clones used in this experiment responded aggressively to contact with the tentacles of all twenty-five non-clonemates but not to contact with a clonemate's tentacle I think it likely that aggression is triggered by contact with any and all non-clonemates.

The following experiment demonstrates that these anemones, while reacting aggressively to contact with the tentacles of any non-clonemate, quite reliably fail to aggress upon contact with the tentacle of a clonemate.

(1) An anemone was stimulated as usual using the excised tentacles of a non-clonemate. Stimulation was discontinued if the reaction was not elicited within ten contacts, and those anemones not reacting within ten contacts were eliminated from the experiment at this point. (2) After a five minute rest period, the anemone was stimulated in exactly the same manner with the tentacle of a clonemate (one kept in a separate bowl to eliminate the possibility of acquired tolerance involving specific individuals). Stimulation was discontinued if the aggressive response was not elicited within twenty contacts. (3) After a five minute recovery period, the anemone was again stimulated with the tentacles of a non-clonemate, and the threshold was recorded.

This experiment was repeated thriteen times using anemones from four different clones. Two animals were eliminated from the experiment after step 1 because they had thresholds greater than ten. Of the remaining eleven animals (Table I) all but one (K) failed to aggress after repeated contact with a clonemate's tentacle. The behavior pattern of anemone K was peculiar in that the agressive behavior was directed ninety degrees away from the area of stimulation, while normally aggression is directed toward the area of stimulation. In this case, aggression was di-

TABLE I

*Inter-clonal specificity in the initiation of the aggressive response. The symbol 7+ indicates
that seven contacts with the excised tentacle initiated the aggressive response in the
experimental animal, while the symbol 20-0 indicates that twenty contacts
with an excised tentacle did not initiate the aggressive response*

Individual anemone	Number of stimulations—response		
	Non-clonemate's tentacles	Clonemate's tentacles	Non-clonemate's tentacles
A	7+	20-0	5+
B	1+	20-0	1+
C	8+	20-0	1+
D	6+	20-0	2+
E	3+	20-0	2+
F	1+	20-0	1+
H	8+	20-0	3+
I	5+	20-0	1+
K	3+	16+	1+
L	5+	20-0	7+
M	1+	20-0	2+

rected toward the source of stimulation during step 1. (It should also be men-
tioned here that there is considerable variation in the behavior of these animals
as there is in the behavior of anemones in general. From time to time and for no
apparent reason a member of this species has failed to react to stimulation that at
another time was quite adequate to elicit a reaction. Sometimes a given anemone
has responded less quickly to stimulation than usual. I have also observed de-
layed movements of application following a few minutes after a usual aggression,
and occasional examples of apparently spontaneous aggression. The response of
anemone K during step 2 of the experiment may have been an example of a de-
layed movement of application, since it was directed toward the area of stimulation
during step 1.) In step 2 of the experiment, all eleven anemones proceeded into
full aggression after less than ten contacts with the tentacle of a non-clonemate,
showing that the animals were still capable of responding aggressively to appro-
priate contact.

Apparently, then, the aggressive response of *A. elegantissima* is elicited by con-
tact with non-clonemates or with any of a number of Anthozoans, but not by con-
tact with clonemates, with hydrozoans, with predator or prey species, or with
inert objects.

Aggression-related damage at interclonal borders. Examination of adjacent
clonal borders in the field has frequently shown aggression and aggression related
damage (white patches of acrorhagial ectoderm on tentacles and column, each
spot surrounded by a ring of necrotic tissue) among the animals at or near these
borders. Aggression is rarely observed among anemones in the midst of a clonal
group.

During low tide anemones were collected from the adjacent borders and from
the middles of two clones whose common interclonal boundary had been observed
to be relatively stable over a one year period. These animals were retracted when

collected and no aggression-related damage was evident; however damage was evident when the anemones expanded after they were placed in sea water in the laboratory. Of the seven anemones collected from the border (3 from one clone and 4 from the other), all showed aggression related damage. Of the nine anemones collected from mid-clone (4 from one clone and 5 from the other) none showed aggression related damage.

Here is evidence that in the field aggression is occurring at the boundaries between adjacent clonal groups and not within clonal groups.

In the laboratory, two adjacent clonal groups that were forced into close proximity eventually developed an anemone-free zone between the two groups (Francis, 1973). During this time numerous aggressive episodes between non-clonemates were observed at the border between the two groups suggesting that aggression was involved in the formation of the anemone-free space between the clones.

Reaction of young anemones to intraspecific contact. Small specimens of *A. elegantissima* were found in a mussel bed at Arroyo Hondo (an area just south of Gaviota, Calif.) in October of 1971. Careful examination showed that the smallest of these (those having 30 or fewer tentacles) had not yet developed acrorhagi. Larger specimens having 30 to 40 tentacles were developing acrorhagi on the adoral side of the verucae closest to the oral disc. It seems reasonable to assume (a) that these incompletely developed anemones were relatively recent products of sexual reproduction, and (b) that they were too young to have undergone asexual binary fission and so were each genetically distinct individuals (non-clonemates).

During the experiments described below I observed interactions between several young anemones without developed acrorhagi. When these animals came into tentacle contact with each other their tentacles twitched, writhed, and retracted from the contact. Often the postures of the animals also changed noticeably after such an encounter, the column becoming longer and thinner. Occasionally I observed movements similar to those of the aggressive response; there were frequently bending and writhing movements of the column, and the edge of the oral disc in the tentacle contact area was sometimes lifted up and back as happens in the aggressive response early in the movement of application (Fig. 4). However, none of these anemones showed the full set of movements in the sequence displayed during the adult aggressive response (*i.e.*, tentacle contact and retraction, column elongation, proximal edge of the oral disc drawn up and back, proximal edge of the oral disc extended out and down). During the 4 such encounters that I watched, one of the anemones (the one that had moved into contact range of the other) moved out of contact range within an hour. Obviously these juveniles lacking acrorhagi were unable to damage each other by the application of acrorhagial ectoderm, and their distribution and response to contact with each other is therefore of interest in ascertaining the relation between the damage inflicted as a result of the aggressive behavior and the clone specific segregation observed for the adults of this species.

Both on the mussels and pebbles on which they were collected and on the glass bowls in which they were kept in the laboratory, these young anemones were usually solitary, that is, out of contact range of each other. When within contact distance they were never observed in passive tentacle to tentacle contact as is common within the clonal groups of this species, but one or both of any proximal

pair was always retracted. Groups of 5 to 9 of the young anemones lacking acrorhagi were placed close together in containers of various sorts in order to make it easy for them to settle close to an another, but these animals always moved apart in from 24 to 48 hours. Apparently then, young specimens of the anemone *A. elegantissima* tend to move in such a way as to eliminate contact with genetically different members of the species, even in the absence of the aggressive response.

Actinea equina, variety *mesembryanthemum*

I observed the north Atlantic intertidal sea anemone *Actinea equina* (var. *mesembryanthemum*) in its natural habitat along the northwestern and southeastern coasts of Scotland and along the southern coast of England. There I found the anemones to be solitary, living out of contact distance with each other. Stephenson, 1935, reports asexual reproduction in the species as rare.

Bonnin (1964) described an aggressive response for this variety of *Actinea equina* (color types green and red) and did some preliminary studies on the specificity of the response which indicated that these anemones more often aggress against animals of the opposite color type than against those of their own color type. In studies that I conducted at the Gatty Marine Laboratory, St. Andrews, Scotland, and the Plymouth Laboratory, Plymouth, England, my findings were entirely to the contrary.

Because many of the anemones that I saw could not clearly be classified as either color type red or color type green, each individual is referred to by a number beside

TABLE II

Specificity of the aggressive response of Actinea equina

Source of the excised tentacles used for stimulation*	Response of 6 different individuals to contact with tentacles excised from 12 different individuals†					
	5R,R'	11r,R	390R,r	220,OG	350G,g	29G,G
5R,R'	O	+	+	+	+	/
11r,R	+	O	+	+	+	+
12r,R	+	+	+	±	+	/
20r,R	+	+	+	+	+	/
19r,r	+	±	+	+	+	+
390R,r	+	+	O	+	+	/
220,OG	+	+	+	O	+	±
350G,g	+	+	+	+	O	+
32g,g*	+	+	±	+	+	
29G,G	+	+	+	+	+	
10G,G	+	+	+	+	+	
41G,G	+	+	+	+	+	

* The numbers each designate a different individual. The first letter(s) following each number indicates the tentacle color of the individual (R = red, r = light red, OR = orangish red, O = orange, OG = orangish green, g = light green, G = green) and the letter(s) following the comma indicates the color of the column (R' = dark red, g = light green with darker green spots).

† The symbol + indicates that stimulation of the anemone elicited an aggressive response within 20 contacts while ± indicates that the anemone failed to respond aggressively to one or two sets of stimulations and responded positively to another set. The symbol O indicates that no aggressive response was elicited. The symbol / indicates that there is no information.

which are letters indicating tentacle color and column color (Table II). Six of the anemones covering the color range from green to red were tested in the usual way by repeated contact with tentacles excised from twelve different animals of various colors; this included 20 contacts with one of their own tentacles. Stimulation was discontinued if an anemone did not respond after 20 stimulations, but the results were not reported as negative unless the animal failed to respond to the stimulation on three different occasions. Tests in which the anemone failed on at least one occasion to respond aggressively to twenty contacts with an excised tentacle, but subsequently responded positively are indicated in Table II, by an underscored +. The results are shown in Table II where each individual is indicated by a number beside which the tentacle color and column color are indicated. The specificity of the response in *Actinea equina* seems to be the same as that in *Anthopleura elegantissima;* the anemones aggress in response to contact with tissue from any genetically distinct individual regardless of color type but not upon contact with tissue from a genetically identical individual (in this case their own tentacle). There was no apparent correlation between color type combinations and the ease with which aggression was elicited.

Anthopleura artemisia

Anthopleura artemisia is found living intertidally along the west coast of America from Alaska to southern California, where it lives attached to rocks and buried in mud, sand, or gravel so that usually only the tentacular crown is extended above the surface (Hand, 1955). I have generally found this species to be solitary, although I have occasionally observed close contact between two animals having similar colors and patterning on the oral disc and tentacles. Since this species is known to reproduce asexually by longitudinal fission (Hand, 1955), such pairs are probably clonemates.

The coloring and patterns of this species are highly variable, and I have never observed close contact between two specimens having distinguishably different color of patterning.

Anthopleura artemisia was observed in the laboratory to aggress in response to contact with non-clonemate members of the same species and in response to contact with its congenerics *A. elegantissima* and *A. xanthogrammica.* The aggressive behavior of *A. artemisia* was identical at least superficially to that described for *A. elegantissima.*

Anthopleura xanthogrammica

According to Hand (1955) *Anthopleura xanthogrammica* "is known from Japan (?) to southern California" (page 53), where the adults are commonly found in pools and crevices just below the mussel beds. In the intertidal near Pacific Grove, California where I have observed and collected these animals I have frequently observed very close contact within groups that could only be non-clonemates since this species does not reproduce asexually.

Anthopleura xanthogrammica has never been observed to display an aggressive response, even when provoked by repeated contact with members of the same species or with its congenerics.

Discussion

The aggressive response of some members of the family Actiniidae is another example of a surprisingly specialized and complex behavior pattern displayed by sea anemones, animals having "primitive" systems for nervous integration and often assumed, therefore, to display a very limited behavioral repertoire. I would like to compare this behavior with some other complex behaviors described for anemones and other coelenterates in order to point out the common as well as the distinctive features of the aggressive behavior.

The aggressive response of *Anthopleura elegantissima, Actinea equina* (Bonnin, 1964), and *Anthopleura artemisia* is like both the raptorial feeding of coelenterates and the shell transfer behavior of some anemones (Ross, 1967) in being, a series of movements directed towards a goal. In addition the conspicuous movements are very similar in the three different kinds of behavior patterns in that they are grossly asymmetrical and directed with respect to the source of the stimulus. The aggressive response includes both symmetrical postural changes that involve the whole animal (see Fig. 3, the elongated, upright posture assumed during aggression) and localized, coordinated movements that are radially asymmetrical and oriented with respect to the source of contact stimulation (*e.g.,* the movement of application, Fig 7). Raptorial feeding in coelenterates could involve symmetrical changes in the condition of the whole animal (*e.g.,* postural changes or changes in the threshold of the nematocysts involved in feeding), however the conspicuous movements are asymmetrical (*e.g.,* movement of the mouth and tentacles) and directed with respect to the area of contact with the prey. Aggression is also comparable, in this regard, to the transfer behavior (movement of the anemones from glass or another surface to a shell) shown by *Calliactis parasitica* (Ross and Sutton, 1961; Ross, 1967), *Actinostola* (Ross, 1967) and *Stomphia coccinea* (Ross, 1967) in response to contact with a shell. The transfer behavior appears to involve symmetrical postural changes in the anemones as well as asymmetrical movements that are oriented with respect to the source of stimulation (*e.g.,* in *Stomphia,* swelling of one side of the pedal disc and movement toward a shell with which the tentacles are in contact).

Aggression might also be compared with the swimming response of coelenterates. In the jellyfish swimming involves rhythmically repetitive, radially symmetrical movements. Directedness is normally achieved not by gross asymmetry in the movements themselves but by subtle and temporary asymmetry in the movements which then leads to changes in the orientation of the whole animal. The same is apparently true for the swimming behavior of the Actinians *Gonactinia, Boloceroides, Boloceractis* and *Bunodiopsis medusoides,* all of which use their tentacles in swimming (Robson, 1966). The case is less clear for the swimming of the anemones *Actinostola* and *Stomphia* (Yentsch and Pierce, 1955; Ross and Sutton, 1967). The behavior here initially involves movements that seem relatively symmetrical and that lead to elongation of the column, expansion of the crown, and finally detachment from the substratum. The swimming movement itself, while apparently producing no effective orientation in the net movement of the anemone, could nonetheless be described as asymmetrically oriented with respect to the source of the stimulus since it begins as alternate bending of the column away from and toward the source of stimulation. However, the orientation of the rhyth-

mic bending then slowly shifts around the column (Robson, 1966), making this swimming movement a difficult one to classify with regard to symmetry and directedness.

It does seem apparent that in some important ways the aggressive behavior is more nearly like raptorial feeding and transfer behavior than it is like swimming; aggression is a directed behavior involving asymmetrical movements that are not repeated rhythmically. However, in some other respects the aggressive response does resemble the swimming behavior of *Stomphia* and *Actinostola*. Both of these behavior patterns involve a specific sequence of movements that, once initiated, continues to completion in the absence of additional or continuing stimulation. In both cases the behavior is initiated by contact with a limited number of species (Ross and Sutton, 1967; and Bonnin, 1964), implying that the initiating stimuli are very specific and presumably chemical in nature.

Several characteristics of the aggressive response are distinctive within the repertoire of anemone behavior. One of these is the peculiar manner in which the response is initiated. For *Anthopleura elegantissima* the initiating stimulus involves repeated contact of the tentacles with some part of a non-clonemate conspecific or some other anthozoan. In describing the aggressive response of *Actinea equina* Bonnin (1964) has noted that a definite time lapse between successive contacts during stimulation seems important in reaching the threshold for the response. Too rapid or massive stimulation usually results in complete retraction of the anemone without eliciting an aggressive response. The same seems to be true for *Antholeura elegantissima*.

The aggressive behavior of these anemones is also remarkable in being a very specialized behavior that functions under very particular and limited circumstances (see also Bonnin, 1964) and that involves the use of specialized movements and equipment. Contact and retraction of the tentacles during initiation of the behavior does not appear to be particularly specialized (the animals respond in a similar way to mild mechanical or electrical stimulation of the tentacles); however some of the movements during the inflation stage of the behavior (*e.g.*, the puffing up of one edge of the capitulum and the associated acrorhagi) and during the movement of application (*e.g.*, the asymmetrical elongation of the oral disc and the downsweeping movement of the inflated acrorhagi) are quite distinctive movements of the aggressive behavior. The acrorhagi are specialized "organs" which have been observed to function only during aggression (Abel, 1954; Bonnin, 1964) and apparently the acrorhagial atrich nematocysts are also used only during aggression.

From the evidence presented on the specificity of the aggressive response of *Anthopleura elegantissima*, it is apparent that the behavior is not directly involved either in defense against predators or capture of prey. Aggression is elicited only by contact with conspecifics and other Anthozoans. Bonnin (1964) found the same to be true for *Actinea equina*.

For *Anthopleura elegantissima* the aggressive response seems to have a clear function in intraspecific interactions. Briefly summarized, the pertinent evidence, presented in this and a previous paper (Francis, 1973) is as follows: (1) Within clonal groups of the aggregating form of *Anthopleura elegantissima* the individuals are in close contact with their neighbors and the tentacles of the adjacent animals

are interlaced. In the field, adjacent clones of these anemones are isolated from each other by anemone-free spaces; and in the laboratory a group of anemones of mixed clonal origins will reaggregate into isolated uniclonal groups (Francis, 1973). (2) The specificity of the aggressive response is directly related to that shown in the segregation and separation phenomena seen both in the laboratory and in the field. Contact between clonemates does not elicit aggression and is commonly seen both in the laboratory and in the field. Contact between non-clonemates elicits aggression and is not maintained between these anemones either in the laboratory or in the field. (3) The response of the victim of an aggressive episode is such as to affect its separation from the non-clonemate aggressor. The damaged anemone may move away from the aggressor, or somtimes it may merely lean away. In the laboratory, anemones that were severely damaged as a result of repeated aggressive episodes have been observed to release their hold on the substratum; in the field, such animals would certainly be swept away in the surf. All of these responses tend to remove the victim from contact with the attacking non-clonemate. (4) The evidence also shows that aggression does occur at the boundary between two clonal groups. In the field, anemones collected from an interclonal border showed aggression-related damage while their clonemates that were collected from the center of the clonal groups showed no such damage. In the laboratory numerous aggressive episodes were observed over a three week period as two clonal groups that had been brought into artificially close proximity interacted at their common boundary (Francis, 1973). This suggests that aggression is important in the formation and maintenance of anemone-free zones between adjacent non-clonemates in this species.

It is possible that the activities of some associated animals such as the chiton *Mopalia muscosa* and the turban snail *Tegula funebralis* are involved in the maintenance, especially of very wide trail-like anemone-free spaces between adjacent clonal aggregations. Field (Biology Department, University of California, Santa Barbara, personal communication in 1969) has found that *Mopalia* is capable of maintaining and extending artificially made clearings in aggregations of *A. elegantissima*; and I have observed the appearance of short anemone-free pockets occupied by *Mopalia* or *Tegula* in previously uninterrupted borders of clonal aggregations.

In addition, the observations and experiments with the pre-aggression juveniles of *A. elegantissima* indicate that the aggressive behavior may not be a necessary condition for clone specific segregation and separation in this species. Contact between genetically different individuals was avoided even in the absence of the aggressive response.

However, the direct effects of the aggressive behavior of *A. elegantissima* would be sufficient to account for clone specific segregation and at least minimal separation between clones; and although other factors may be involved, the aggressive response undoubtedly functions as part of an intraspecific territorial behavior in this species.

Assuming that the aggressive response of acrorhagus-bearing members of the family Actiniidae is monophyletic in orgin, and knowing that no function outside of aggression is known for the acrorhagi (Äbel, 1954), we may speculate on the evolutionary advantages and history of this highly specialized behavior.

I have shown that aggression is effective in intraspecific interactions for *Anthopleura elegantissima*. In *Actinea equina* also the specificity of the aggressive response correlates with the intraspecific associations apparent in the field; these anemones are solitary and they respond aggressively to contact with any other conspecific. For *Anthopleura artemisia* the data are less complete since no extensive work has been done on the intraspecific specifiicity of aggression for this species. Nonetheless these anemones do respond aggressively to contact with conspecifics, and they have not been found living in contact with genetically different conspecifics. *Anthopleura xanthogrammica* apparently lacks an aggressive response while having the specialized equipment (acrorhagi bearing atrich nematocysts) used in this behavior. However, the relationship between distribution and behavior is also apparent for *A. xanthogrammica* since genetically different members of this species are frequently found living in close contact, with the tentacles of adjacent non-clonemates passively interlaced. Taken together, this information suggests that the aggressive response of acrorhagus-bearing anemones is presently effective in intraspecific interactions.

This suggests the possibility that aggression arose in this group of animals through intraspecific interactions. If this were true, what might be the origin of and selective advantage(s) for intraspecific territoriality involving mutal repulsion among genetically unlike members of a species? I would suggest that an anemone that repulses all members of the species except clonemates may gain space on the substratum for its genotype. This could be a decided advantage to the genotype possessing this characteristic, especially if there were competition among members of the species for space itself or for the available food in a given area. As long as the adverse effects of this characteristic, such as the inability to form inter-clonal aggregations and thereby reduce water loss (Roberts, 1941) and the energy loss involved in the repulsion of non-clonemates, were not too great any anemone that developed some form of genetically determined interclonal "repulsiveness" would have a competitive advantage over its inoffensive neighbors. This genotype and the post-larval progeny inheriting the characteristic would be assured of adequate "living space" and would therefore have an increased probability of survival and reproductive success. It may have been in this way that the evolutionary precursor of the aggressive response (perhaps a simple tendency to avoid contact, as in the case of the pre-aggression young of *Anthopleura elegantissima*) became fixed in the common ancestor of these anemones.

It seems apparent to me that an aggressive behavior that arose through intraspecific competition would be of value only to the individual anemones that display the behavior and not to the species as a whole. (Of course a characteristic theoretically need not be of advantage to the species in order to be selected for, because selection is believed to operate at the level of the individual.) Once aggression and armament are adopted and convey their temporary advantage to the aggressor, an evolutionary trend toward more potent weapons and more effective ways of using them seems very likely. Among the acrorhagus-bearing anemones the specialized nematocysts carried on specialized protuberances (the acrorhagi) and the relatively effective behavior pattern that brings these weapons into play may have evolved in this way.

It is also possible that the aggressive response evolved through interspecific

interactions with other anthozoans and served to counter direct competition for space and food by other large sessile coelenterates. This hypothesis is appealing in that it provides a selective advantage to the species as well as to the individual in the origin and development of an aggressive response. However, while intraspecific contact is effective at eliciting aggression in all three species for which the response has been described, interspecific contact is a less reliable elicitor than intraspecific contact for one of the two species tested in this regard. Both interspecific and intraspecific contact are reported to be completely effective at eliciting aggression in *Actinea equina* (Bonnin, 1964), while intraspecific contact is much more reliable than interspecific contact at eliciting aggression in *Anthopleura elegantissima*. For *Anthopleura xanthogrammica* both the interspecific and the intraspecific components of the response have apparently been lost (presumably in recent time since the supposedly vestigal acrorhagi do not appear to be different in form or number from the functional ones of *A. elegantissima*).

Before this question of evolution can be dealt with further, we clearly need more information, especially on the interspecific aspects of the aggressive response of acrorhagus-bearing Actiniids.

Summary

Tentacle contact between genetically different individuals of the west coast sea anemone *Anthopleura elegantissima* initiates in one or both individuals an elaborate behavior pattern that usually results in damage to one or both animals. This aggressive behavior may be described as progressing through five stages: (1) stimulation (or initiation) during which the tentacles of the two anemones repeatedly come into contact and then withdraw from contact; (2) inflation, during which the acrorhagi commence to become turgid; (3) the movement of application, during which the inflated acrorhagi are pushed toward the source of stimulation; (4) application of ectoderm, during which damage is inflicted on the other anemone by the application of acrorhagial ectoderm to the body of the victim; and (5) recovery, during which the anemone returns to its normal posture.

The tissue of the victim to which acrorhagial ectoderm has been applied by an aggressor becomes necrotic and sloughs off within a few days. Sections of such tissue that were fixed and sectioned immediately after the application of ectoderm showed penetration of the tissue of the victim by the large atrich nematocysts in the aggressor's acrorhagial ectoderm.

The aggressive response of *Anthopleura elegantissima* is like raptorial feeding and the shell transfer behavior of some anemones in that it is a directed behavior involving asymmetrical movements that are not repeated rhythmically. Aggression resembles the swimming behavior of *Stomphia* and *Actinostola* in that the specific

sequence of movements, once initiated, continues to completion in the absence of additional or continuing stimulation. The deployment of specialized effector "organs" (the acrorhagi) bearing specialized nematocysts (the large acrorhagial atrichs), the necessity for repeated contact in initiating the response, and the functioning of aggression in intraspecific territoriality are interesting characteristics of this behavior that are unique in the repertoire of anemone behavior.

For *Anthopleura elegantissima* the aggressive response is not directly involved either in defense against predators or capture of prey. Evidence presented here suggests that for this species the aggressive response is involved in intraspecific interaction. Pertinent evidence, presented in this and a previous paper (Francis, 1973) is as follows: (1) Within clonal groups of the aggregating form of *Anthopleura elegantissima* the individuals are in close contact with their neighbors and the tentacles of the adjacent animals are interlaced. In the field, adjacent clones of these anemones are isolated from each other by anemone-free spaces; and in the laboratory a group of anemones of mixed clonal origins will reaggregate into isolated uniclonal groups (Francis, 1973a). (2) The specificity of the aggressive response is directly related to that shown in the segregation and separation phenomena seen both in the laboratory and in the field. Contact between clonemates does not elicit aggression and is commonly seen both in the laboratory and in the field. Contact between non-clonemates elicits aggression and is not maintained between these anemones either in the laboratory or in the field. (3) The response of the victim of an aggressive episode is such as to affect its separation from the non-clonemate aggressor. The damaged anemone may move away from the aggressor, or sometimes it may merely lean away. In the laboratory, anemones that were severely damaged as a result of repeated aggressive episodes have been observed to relase their hold on the substratum; in the field, such animals would certainly be swept away in the surf. (4) The evidence also shows that aggression does occur at the boundary between two clonal groups. In the field, anemones collected from an interclonal border showed aggression-related damage while their clonemates that were collected from the center of the clonal groups showed no such damage. In the laboratory numerous aggressive episodes were observed over a three week period as two clonal groups that had been brought into artificially close proximity interacted at their common boundary (Francis, 1973a). This suggests that aggression is important in the formation and maintenance of anemone-free zones between adjacent non-clonemates in this species.

The direct effects of the aggressive behavior of *A. elegantissima* would be sufficient to account for clone specific segregation and at least minimal separation between clones; and although other factors, such as the activities of associated animals and the tendency of the young anemones to move apart even in the absence of the aggressive response, may be involved, the aggressive response undoubtedly functions as part of an intraspecific territorial behavior in this species.

In *Actinea equina* also the specificity of the aggressive response correlates with the intraspecific associations apparent in the field; these anemones are solitary and they respond aggressively to contact with any other conspecific. No work has been done on the specificity of the aggressive response in *Anthopleura artemisia,* however these anemones do respond aggressively to contact with conspecifics, and they have not been found living in contact with genetically different conspecifics.

Anthopleura xanthogrammica apparently lacks an aggressive response while having the specialized equipment (acrorhagi bearing atrich nematocysts) used in this behavior. However, the relationship between distribution and behavior is also apparent for *A. xanthogrammica* since genetically different members of this species are frequently found living in close contact, with the tentacles of adjacent non-clonemates passively interlaced. Taken together, this information suggests that the aggressive response of acrorhagus-bearing anemones is presently effective in intraspecific interactions.

It is suggested that the evolutionary origin of the aggressive response among acrorhagus-bearing members of the family Actiniidae is monophyletic and that the behavior may have arisen in connection with competition for space either within the species of with other anthozoan species.

LITERATURE CITED

ÄBEL E. F., 1954. Ein Beitrag zur Giftwirkung der Actinien und Function der Randsäckchen. *Zool. Anz.*, **153** : 259–268.

BONNIN. J.-P., 1964. Recherches sur la "reaction d'aggression" et sur le fonctionnement des acrorhages d'*Actinia equina* L. *Bull. Biol. Fr. Belg.*, **1** : 225–250.

CARTHY, J. D., AND F. J. EBLING, Eds., 1964. *The Natural History of Aggression*. Institute of Biology Symposium No. 13, Academic Press, New York, 449 pp.

FRANCIS, L., 1973. Clone specific segregation in the sea anemone *Anthopleura elegantissima*. *Biol. Bull.*, **144** : 64–72.

HAND, C., 1955. The sea anemones of central California. Part II. The Endomyarian and Mesomyarian anemones. *Wasman J. Biol.*, **13** : 37–97.

ROBERTS, B. J., 1941. A survey of the methods employed by intertidal organisms in resisting desiccation. *Masters thesis, Stanford University*, 68 pp.

ROBSON, E. A., 1966. Swimming in Actiniaria. *Symp. Zool. Soc. London*, **16** : 333–360.

ROSS, D. M., 1967. Behavioral and ecological relationships between sea anemones and other invertebrates. *Oceanogr. Mar. Biol. Annu. Rev.*, **5** : 291–316.

ROSS, D. M., AND L. SUTTON, 1961. The response of the sea anemone *Calliactis parasitica* to the shell of the hermit crab *Pagurus bernhardus*. *Proc. Roy. Soc. London, Series B*, **155**(959) : 266–281.

ROSS, D. M., AND L. SUTTON, 1967. Swimming sea anemones of Puget Sound: Swimming of *Actinostola New Species* in Response to *Stomphia coccinea*. *Science*, **155** : 1419–1421.

STEPHENSON, T. A., 1935. *The British Sea Anemones, Vol. 2*. Ray Society, Dualau and Co., London, 142 pp.

YENTSCH, C. S., AND D. C. PIERCE, 1955. "Swimming" anemones from Puget Sound. *Science*, 122 : 1231–1233.